PHYLLOTAXIC DIVERSITY IN *MAGNOLIA* FLOWERS

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ABSTRACT

Impermanent initials and thus the shift of the axis in the stochastic meristems are postulated to be responsible for ontogenetic phyllotactic diversity in plants. In the gynoeceum of *Magnolia acuminata* the main Fibonacci pattern develops in much less than 50% of individual generative shoots. There is also an extremely wide spectrum of other patterns, among them even the rarest 1, 3, 8, 11... pattern is present. Regarded sometimes as "impossible", the pattern has been documented in SEM for the first time. Beside the presence of various patterns, frequent ontogenetic transformations of phyllotaxis have been found in *Magnolia*. These are indicated by dislocations in the periodic distribution of carpel primordia. In other magnolias, exemplified by *M. soulangeana*, the Fibonacci pattern prevails, but not as much as in coniferous vegetative shoots, where, as demonstrated earlier, it reaches 95%. Other pattern numbers are also different. This suggests the involvement of the genetic factor and may be attributed to the higher frequency of discontinuous phyllotactic transformations in some species. The stochastic character of the meristem is perhaps more pronounced in some plants, which leads in turn to more frequent transitions and greater pattern diversity.

KEY WORDS: *Magnolia*, phyllotaxis, crystal lattice, apical meristem, dislocations, plant morphogenesis.

INTRODUCTION

Regularity in the distribution of such repetitive elements of the plant body as leaves or flower parts, known as phyllotaxis, has been for a long time one of the most intriguing phenomena in plant science. Our knowledge is still insufficient in understanding how the regularity is accomplished. Even less is known about why phyllotaxis has so many images.

Various phyllotactic patterns have been documented in botanical literature as characteristic of different plant species and different plant structures (Fujita 1937, 1939, Zagórska-Marek 1985, Jean 1992, 1994). Among them the distichous and whorled patterns are distinguished where such pattern elements as leaves, flowers or flower parts can be connected with vertical lines, called ortostichies. In alternate patterns the connecting lines – parastichies, assume either a spiral or helical course depending on a geometric form of a structure in which the pattern is studied. In the first case it is a paraboloid dome of the apical meristem, in the second, a cylindrical surface of the elongated stem. Alternate phyllotaxis is therefore often termed as spiral or helical. The parastichies in opposite sets, winding up in opposite directions: clockwise and counterclockwise, are inclined at different angles to the stem axis and their number in both sets is not the same: 2 and 3 or 3 and 4. Thus the distinct feature of this phyllotaxis is chirality – each pattern may exist in two mirror-like images, two chiral configurations. In whorled phyllotaxis the parastichies are also present, but in opposite sets there is an equal number of them: 2:2 or 3:3. Although opposite, they are inclined at the same angle to the axis of the shoot. This phyllotaxis is not

chiral. The overview of phyllotactic patterns occurring in nature is given in Table 1.

In plants with alternate phyllotaxis one of the patterns is most common. It is the main Fibonacci pattern in which the numbers of possible parastichies and possible connections between leaves along parastichies, make the famous sequence of numbers known as Fibonacci series: 1, 1, 2, 3, 5, 8, 13.... Other alternate patterns (Table 1) have been noted by some investigators as occurring very seldom, almost accidentally and inconsistently in various plant species otherwise characterized by the main Fibonacci pattern. Being rare and unpredictable they seemed to represent rather the puzzling fantasy of nature than the instructive and enlightening source of knowledge on the mechanism governing phyllotactic pattern formation. Phyllotaxis research is very clearly divided into empirical and mathematically oriented. Some of the theoretical models do not necessarily coincide with reality. In one extreme case the very existence of some patterns such as 1, 3, 7, 10.... or 1, 3, 8, 11... and consequently the credibility of the authors reporting them, has been questioned by a theoretician considering these patterns impossible (Jean 1992, 1994).

A few years ago the results of studies on *Abies balsamea* vegetative shoots showed, that in one species only and in one type of plant structure as many as twelve different patterns of alternate phyllotaxis may be encountered (Zagórska-Marek 1985). The most important finding however was, that one alternate pattern may transform ontogenetically into another, also alternate. It is interesting, that the change of the pattern in *Abies* appeared to obey some principles. It minimized the difference in parastichy number and inclination, between the

TABLE 1. Natural patterns of phyllotaxis encountered in plants.

I. Alternate patterns:			
1. Monojugate (simple) patterns with a single ontogenetic helix:			
	<u>Divergence</u>	<u>Parastichy numbers</u>	<u>Name of the pattern</u>
	137.5	1, 1, 2, 3, 5, 8, 13, 21...	main Fibonacci
	132.2	1, 3, 8, 11, 19, 30...	
	99.5	1, 3, 4, 7, 11, 18, 29...	Lucas
	106.4	1, 3, 7, 10, 17, 27...	
	77.9	1, 4, 5, 9, 14, 23...	
	64.1	1, 5, 6, 11, 17, 28...	
	151.1	1, 2, 5, 7, 12, 19, 31...	
	158.1	1, 2, 7, 9, 16, 25...	
2. Polijugate (multiple) patterns with more than one ontogenetic helix (j):			
j = 2			
	137.5/2	2, 4, 6, 10, 16, 26...	main Fibonacci bijugy
	99.5/2	2, 6, 8, 14, 22, 36...	Lucas bijugy
j = 3			
	137.5/3	3, 6, 9, 15, 24, 39...	main Fibonacci trijugy
j = 4			
	137.5/4	4, 8, 12, 20, 32...	main Fibonacci tetrajugy
II. Whorled patterns with k number of elements in one whorl:			
<u>Circumferential distance between whorl elements(°)</u>	<u>Parastichy numbers</u>	<u>Ortostichy numbers</u>	<u>Name of the pattern</u>
k =1			
360	1:1	2	distichous
k=2			
180	2:2	4	decussate
k=3			
120	3:3	6	tricussate
k=4			
90	4:4	8	tetracussate
k= 5, 6, 7, 8....

Fig. 1. Flowers of *Magnolia*. Their hidden beauty is the astoundingly diverse phyllotaxis of gynoecia.

Fig. 3. Generative shoot of *Magnolia* at the end of blooming. In the first step the perianth falls off, then the anthers and the carpel styles dry out. Gynoecium expands and its spirally arranged carpels start maturing. This is the stage when the phyllotaxis of this structure can be easily determined.

with the imperfections of the atomic or the molecular network of crystal known to crystallographers as dislocations (Zagórska-Marek 1987). Their meaning in plants is such that there is a possibility of transformation from one pattern to another between all patterns of phyllotaxis. This discovery undermines some classic descriptive criteria used by the students of phyllotaxis. The divergence angle and ontogenetic helix become nothing but useful abstracts since in many transitions only one parastichy missing from the lattice at the site of the dislocation, means a dramatic change not only in the divergence angle but also in the direction of the ontogenetic helix, i.e. in the chirality of initiation.

A new interpretation of phyllotactic diversity has been proposed on the basis of these findings. It has been postulated that phyllotactic pattern is created in a manner resembling crystal growth. In the system of equally spaced primordia, the unit of the pattern (PTU) has been defined as a common element of all phyllotactic patterns (Zagórska-Marek 1987). According to this hypothesis the sectorial i.e. discontinuous change in the apex circumference introduces a dislocation and thus alters phyllotaxis. Every imaginable pattern can be created from any initial pattern through such local instability of the apex circumference as it can be seen from the grid made of PTU (Fig. 2). The quality of the pattern depends on the ratio between the sizes of primordia and apex circumference and upon the inclination of PTU units in the grid with respect to the vertical axis of the meristem. The inclination of the lat-

ter has to change with the transformation of phyllotaxis. The most important is, that the PTU hypothesis offers certain unification of all phyllotactic patterns – there is basically no difference between alternate and whorled phyllotaxis since they easily may transform one into another in a following manner: $2:2 \rightarrow 2:3$; $3:3 \rightarrow 3:4$; $4:4 \rightarrow 4:5$ etc. Indeed, the ontogenetic sequence of whorled and spiral phyllotactic patterns recently described in *Anagallis* is exactly the same as modelled on the grid assuming that the principle of minimal axial and circumferential change is observed. In *Anagallis* the initial decussate $2:2$ pattern transforms into the Fibonacci $2:3$ then into tricusate $3:3$ and finally into the Lucas pattern $3:4$ (Kwiatkowska 1993, 1994). The sequence indicates either ontogenetic decrease of the primordia size relative to the apex circumference or ontogenetic increase of the latter relative to the primordia size.

The ontogenetic transformation of the decussate (whorled) phyllotaxis into spiral often occurs in the advanced ontogeny of plants such as *Bryophyllum* (Jensen 1968, Gómez-Campo 1974) or *Anagallis* (Kwiatkowska 1994). Since all Dicotyledones have opposite first leaves (cotyledones) and many Gymnosperms such as *Abies* or *Picea* always have opposite prophylls in axillary buds (Romberger et al. 1993), we must regard this type of transition as the most common. The reverse situation is frequent in many plants at the onset of flowering. Recently it has been described in *Impatiens balsamina*. In this plant during the transition to flowering, helical

Fig. 4. *Magnolia acuminata* – elongated, mature gynoecia with the characteristic helical arrangement of carpels on the lateral surface of the cylindrical shoot. The parastichies winding upward either divide (A) or fuse (B). The change in the number of parastichies signalizes an ontogenetic transformation of phyllotaxis. The arrow points out the vertex of parastichy dislocation. The type of transition in a particular shoot is given at the bottom of each photograph. The initial tetrajugy changes into the main Fibonacci pattern in A, and into the Lucas pattern in B. In A, the upper portion of the gynoecium is frost damaged.

phyllotaxis of the vegetative shoot changes ontogenetically into whorled, which is typical of floral structures. It is interesting, that the reversion of the floral apex already producing a whorled pattern, back to the vegetative stage, does not alter phyllotaxis. In reverted shoots leaves are in whorls i.e. according to a pattern atypical for the plant's vegetative stage (Battey et al. 1993). The *Impatiens* case as well as transitions in coniferous vegetative shoots, show clearly, that it is not necessarily a change in the size or quality of primordia (floral, vegetative) that facilitates phyllotactic transition. This might indicate that the most important factor inducing transition is the sectorial instability of the apex circumference.

Alternate patterns other than the main Fibonacci are rare, so are their transformations. Only 2 – 5% of vegetative shoots in conifers have a pattern differing from the Fibonacci (Gregory and Romberger 1972, Zagórska-Marek 1985, Jean 1992, 1994). This frequency is insufficient to study the relationships between various parameters of apex geometry such as the size of primordia versus apex circumference or the position of the vertex in different patterns. Such measurements are necessary to test the hypothesis on discontinuous, sectorial changes of apex circumference. Being infrequent neither the patterns themselves nor the dislocations were properly documented. Fortunately the perfect object has been found – it is the mag-

nolia gynoecium where many apocarpic carpels are spirally arranged on the elongated floral axis (Fig. 1, 3). The diversity of patterns is here unexpectedly rich and phyllotactic transitions are frequent. The cause of this unusual diversity is not known. The aim of this paper is to present the phenomenon, to advertise the magnolia generative shoot as exceptionally suitable for phyllotactic research and especially to demonstrate the „impossible” patterns.

MATERIAL AND METHODS

Magnolia gynoecia have been collected and analysed since 1985 when a small tree of *Magnolia soulangeana* growing on the lawn of Harvard Forest (Petersham, Massachusetts, USA) was examined by the author of this work. Variant phyllotactic patterns in the gynoecia were then noticed for the first time. *Magnolia*'s generative shoots were usually collected, especially for quantitative studies, in the form of mature cylindrical structures (green „cucumbers”) that remain on the tree after flowering (Fig 3). The number of gathered shoots depended on the yield of an individual tree in a particular season and also on the accessibility of the shoots, which not always fall off the trees. For comparison six different species of *Magnolia* had been examined. These were: *M. acuminata*, *M.*

denudata, *M. salicifolia*, *M. soulangeana*, *M. stellata* and *M. virginiana*. Each species was represented by more than one individual tree. If possible, trees of the same species growing in geographically different locations were studied. These were: The Botanical Garden of Wrocław University in Poland, Harvard Forest in Petersham, Massachusetts and The Miami University Campus in Oxford, Ohio in USA.

To determine the phyllotactic pattern, the parastichies winding in opposite directions were counted at the base and again near the top of each gynoecium to make sure that any transition would not go unnoticed. Contact parastichies were usually selected for counting. These are the most conspicuous lines since they intersect each other nearly at a right angle. The orientation of the parastichies in two opposite sets was indicated by the letters S (ascending clockwise) and Z (ascending counterclockwise) accompanying the numbers in the opposed parastichy pair formula. By means of extrapolating the numbers into a phyllotactic series and reading the chiral configuration of the ontogenetic helix – the overall chirality of each pattern had been determined. The most interesting cases of patterns or transformations were recorded by rolling the gynoecia over the surface of modelling clay. The impressions left by their lateral surfaces were then photographed.

The analysis of mature, elongated gynoecia was supplemented by examining the embryonic structures of *Magnolia* flowers in SEM. Floral buds were collected at the time of laying down carpel primordia i.e. between June and July. To prepare the material for observation in SEM the method of resin casting has been used. Apices after removing all outer parts (stipules and tepals) were replicated in hydrophilic dental impression plastic (Reprosil Caulk Dentsply, USA). Polymerized plastic molds were filled with liquid epoxy resin. After hardening the epoxy casts of apices were sputter-coated and then analysed and photographed in SEM. Sometimes, if the hardening was insufficient, due to some undetermined resin properties, a very fine pattern of cells recorded from the surface of the apex, melted in the sputter-coater and disappeared from the surface of the cast. On SEM micrographs of the floral apex taken from above, the parastichies are seen as spirals along which the carpel primordia are lined up. Pattern identification proceeded by recording the number of spirals, similarly as on mature gynoecia.

RESULTS

In elongated gynoecia of the examined species of *Magnolia* (Fig. 4) there was no less than 50 carpels making one shoot. This was quite a sufficient number to make phyllotactic pattern identification possible and accurate. Numbers of contact parastichy pairs varied greatly from one shoot to another, especially in *M. acuminata*, indicating significant phyllotactic diversity (Fig. 5). Nevertheless the expressions of the patterns, i.e. contact parastichy numbers, were similar in different shoots. Such classically different patterns as multiple and simple (see Table 1) were expressed by the numbers 6:9, 6:8 and 6:7, by 6:10 and 7:10 or, as in Figs 4, 5, by 4:8 and 4:7. The expression of the phyllotactic pattern depends on the size of the pattern elements in relation to the circumference of the shoot. The same pattern, such as main Fibonacci or Lucas, can be expressed differently depending on contact parastichy numbers. In small shoots their expression will be 3:5 and 3:4, whereas in bigger shoots the numbers will be higher: 8:13 and 7:11. In *Magnolia* each pattern, without exception, was found to occur in both chiral configurations (Fig. 6). They were almost equally frequent. Phyllotactic transformations marked by

the presence of dislocations were easy to notice in elongated gynoecia: along the axis of gynoecium parastichies often multiplied or disappeared from the sets (Figs 4, 7). Transformations based on the number of parastichies ontogenetically reduced clearly prevailed (Fig. 7).

The rich diversity of phyllotactic patterns and the presence of dislocations observed on elongated maturing gynoecia has been confirmed by SEM analysis of the embryonic stages of pattern formation (Figs 8, 10-14). Therefore the diversity represents an original feature of the investigated structure and is not merely a product of the initial pattern being distorted by growth and differentiation. In *Magnolia* floral apices the carpel primordia arise as significantly larger structures above smaller stamens (Fig. 8A). They are usually round and discrete because of deep fissures between neighboring primordia. Sometimes atop of a dislocation, where two parastichy joined one another, the primordia fusion was observed (Figs 8, 9).

Almost all known phyllotactic patterns had been recorded including the rare, although undoubtedly existent, patterns 1, 3, 8, 11... (Fig. 10). and 1, 3, 7, 10... (Fig. 11). The comparison of their SEM images with the image of a very frequent in *Magnolia* trijugate pattern 3, 6, 9, 15... (Fig. 12) shows, that geometrically all these three patterns are not very different one from another. Clear dislocations (Fig. 13) or uppermost disturbances in primordia sequence signalling ongoing transformation (Fig. 14) were frequently observed and had been recorded on SEM pictures.

Alternate patterns prevailed significantly. Whorled patterns were also encountered, but only sporadically and only in *M. acuminata*. They were usually of high order i.e. with a high number of parastichies in each set. The expression of such patterns was similar to that noted by Davis and Bose (1971) in the spadices of some aroids: 6:6, 7:7 or 8:8. In alternate patterns the numbers of contact parastichies were: 6:8, 6:7, 6:9, 8:12, 8:11, 7:11, 7:10. This means that at the time of initiation the order of phyllotaxis in *Magnolia* is high and the size of the carpel primordia relative to the apex circumference is small. The expression of phyllotaxis may change with the elongation of the gynoecia. For example it was noticed, that tetrajugy expressed in elongated gynoecia by the numbers 4:8, at the embryonic stage of flower development was usually of higher order i.e. 8:12. Similarly the Lucas pattern was expressed as 4:7 and 7:11, while some simple accessory patterns as 3:7 and 7:10 or 3:8 and 8:11 (for comparison see Figs 6 and 11 or Figs 7 and 10). Simulation of an identical effect of lowering of the order of phyllotaxis with the elongation of the shoot was made for coniferous shoots by Cannel and Bowler (1978). However, the change in the expression of phyllotaxis in the elongating gynoecium is not always the case, since during differentiation it not only elongates, but also grows in circumference. The ultimate effect, i.e. the decrease in the order of phyllotaxis, depends upon how synchronized the changes in these two parameters of growth are during maturation. For that very reason the expression of phyllotaxis in elongated gynoecia was regarded as less significant than that determined in floral meristems.

As detected on the basis of examining the mature gynoecia, different species of *Magnolia* varied significantly in the frequency of phyllotactic patterns. In *M. virginiana* phyllotaxis was very uniform. Among 141 shoots collected from four trees only two had a pattern different from the main Fibonacci. These were the 5z:7s and 5s:9z patterns, both from the same tree. In *M. stellata*, *M. salicifolia* and *M. soulangeana* the Fibonacci pattern clearly prevailed (Fig. 15). The greatest diversity was found in *M. acuminata* in every case (Fig. 16).

Fig. 5. *Magnolia acuminata* – pure phyllotactic patterns recorded by pressing and rolling the mature gynoecia over the surface of modelling clay. An interesting illusion, which depends upon the direction of light, allows the perception of the pattern depressions (made by ovaries) as elevated structures. This simulates the sculpture of the lateral surface in a real gynoecium (see Fig. 4). The patterns of unrolled phyllotactic helices resemble crystal lattices. The number of helices in each set of the opposed parastichies, thus the type of the pattern, is easy to determine since some ovaries are replicated once again on the opposite side of the clay impression, when the shoot is rolled for more than 360 degrees. They are recognizable on the basis of minute characteristic details of their structure, which are well recorded in the impression. Such ovaries serve as the surface marking points (black dots). Pattern identification is given in the bottom left corner of each picture. The letters Z and S accompanying the numbers of the opposed parastichy pair indicate the orientation of parastichies on the replica. It is reverse to that existing in the shoot, because the chirality of the replicated pattern is always a mirror-image of the real one – we look at it as from the inside of the shoot.

Fig. 6. *Magnolia acuminata* – modelling clay impressions of two different shoots in which the rare 1, 3, 7, 10... pattern was present. It was very regular in both cases. The chirality of ontogenetic helix in one shoot was Z (upper photograph) and opposite in the second. As explained earlier, the chirality of the recorded pattern is a mirror-image of the real one. Further description as in Fig. 5. The length of the bar is 5 mm.

There was however one individual tree of this species, which had a pattern of phyllotactic diversity more similar to those in the other examined species. The main Fibonacci pattern in the gynoecia of this tree was dominant.

The most interesting is the evidence of gradual increase of diversity among the investigated species. It is the lowest in the case of *M. soulangeana* and *M. salicifolia*, slightly higher in *M. stellata* and quite well pronounced in *M. denudata*, where distribution and frequency of patterns resembled that of described earlier, exceptional tree of *M. acuminata*.

The quantitative analysis of the transition frequency showed that it was greater in trees with a wider spectrum of phyllotactic patterns. In such trees the number of shoots with irregular phyllotaxis was also big. Species with a high frequency of the Fibonacci pattern had a smaller number of shoots with dislocations. The preliminary study showed that in *M. acuminata* some transformations were more numerous than others (Fig. 17). There were also differences in this respect between individual trees. In one of them the most frequent was the transformation of the 7:10 pattern into 6:10. In two others the transformation of 4:8 into 4:7 and of 6:8 into 5:8. The significance of this observation has not yet been esti-

mated statistically. The phenomenon could be dependent on the frequency of the initial pattern later being altered. On the other hand, the geometric stability of the initial pattern can not be excluded as the factor inducing its own transformation.

It should be emphasized that the natural transformations found in *Magnolia* usually involved the patterns located within the same domain of the phyllotactic grid. For instance the 3:7 pattern transformed into patterns such as 3:6, 4:6, 2:7 and 4:7. Alteration of the main Fibonacci pattern 5:8 usually resulted in 5:7, 4:8, 6:8, 4:7, 6:7, 6:9 patterns. Yet another pattern 3:8 produced 4:8, 4:7, 2:7, 3:7 or 4:6 patterns, which means that in all these transformations both the axial and circumferential changes, as read from the phyllotactic grid, were minimal.

DISCUSSION

Phyllotaxis of the vegetative shoots in *Magnolia* is either spiral or distichous (Charlton 1994). The phyllotactic continuum has not been studied in this plant, thus we do not know how the vegetative phyllotaxis might affect the ultimate pattern of the flower.

The taxonomists studying the development and morphology of the flower in *Magnoliaceae* (Tucker 1960, 1961, Erbar and Leins 1982, 1983, Nooteboom 1985, 1987, 1993) agree that perianth elements, if spiral as it is in *Magnolia stellata*, are initiated in one spiral, whereas stamens and carpels are in 8 (7-10) spirals. This is not an entirely correct notation since in the compact system of stamens and carpels, the basic spiral (ontogenetic) also exists, but is inconspicuous. Moreover, there is no reason for selecting from both numbers of contact parastichies only one. What correctly defines the phyllotactic pattern is the opposed parastichy pair. Therefore two numbers of spirals: oriented to the left and to the right have to be given. Eight spirals may belong to the main Fibonacci series but also to the series 1, 3, 8, 11... Thus there are at least two different patterns represented by eight spirals. Seven spirals most likely represent the Lucas pattern but the number seven can also be found in at least three other phyllotactic series (see Table 1). In his "Notes on *Magnoliaceae*" Nooteboom writes about the dilemma of changing orientation of the basal spiral and about "too early" development of some stamens: 9-th before 6-th, 17-th before 14-th and 25-th before 22-nd, the phenomenon described as „some androecium members hurrying on in advance" by Erbar and Leins (1982, 1983). These facts are easy to explain in the light of the "crystalline" concept of phyllotaxis promoted in this paper. The reversal of ontogenetic helix as well as sectorial irregularities in primordia distribution are commonly associated with dislocations and phyllotactic transformations. In *Abies balsamea* the leaves supposedly older were often located along the ontogenetic helix above the younger (Zagórska-Marek 1985, 1987). Despite that, the pattern of high order parastichies was always remarkably regular. Having noted in the floral apices of *Magnolia*, numbers of spirals different than those in the most common phyllotactic series, such as 7 – 10, the other authors did not define there other patterns, than the main Fibonacci.

The high frequency of various phyllotactic patterns and dislocations in *Magnolia*, reported in this work, was found not only in the elongated gynoecia but also in the SEM examined floral apices representing the early stages of phyllotactic pattern formation. This excludes the possibility of pattern distortion in a course of gynoecium growth and differentiation. Thus, an inaccurate reading of phyllotaxis from the elongated

Fig. 7. *Magnolia acuminata* – phyllotactic transitions recorded in modelling clay. The imperfections of lattices known in crystallography as dislocations are indicators of the transitions. They are shown with heavy lines. In the upper right shoot two dislocations occur independently in two different locations of its lateral surface. In the upper left shoot two ovaries situated atop of the dislocation join one another. In the middle right shoot the enlarged ovary "embraces" two parastichies at the point, where one of them ends its course. Further description as in Fig. 5.

Fig. 8. SEM images of the epoxy casts of *Magnolia* floral apices, replicated at the stage of laying down carpel (c) and stamen (s) primordia. A, B – *Magnolia soulangeana*. C, D – *Magnolia acuminata*. At a higher magnification it can be appreciated how well the epoxy cast preserves the cellular pattern of the apex. It is because of the hydrophilic, nontoxic properties of Reprosil, the intermediate medium. In the first apex (A, B) a very regular main Fibonacci pattern is present, expressed by 5z:8s contact parastichy numbers. In the second apex (C, D) phyllotaxis changes, which is indicated by a clear dislocation in a course of parastichies oriented to the left (shown with broken lines). At the vertex of the dislocation, marked by an arrow, the carpel primordia fuse. From this level upward only one parastichy runs instead of two. The transition present in this apex is thoroughly analyzed on Fig. 13. As can be seen on A, stamen primordia, although initiated earlier and thus advanced in their development, are smaller than the carpel primordia. The change in size itself can not be attributed to the phenomenon of phyllotactic transition since the transitions predominantly occur not between the androecium and gynoecium but within the gynoecium, well above the border between the stamen and the carpel zone. The bar length is 100 μm on A, C and 50 μm on B, D.

Fig. 9. *Magnolia acuminata* – another example of primordia fusion shown magnified in the lateral view of the apex organogenic region. The length of the bar is 50 μm .

Fig. 10. *Magnolia acuminata* – the exotic, extremely rare pattern of 1, 3, 8, 11.... The divergence in this pattern is 132,36 degrees, very close to the golden angle of the main Fibonacci pattern (137,5). Parastichies being the first members of the Fibonacci phyllotactic series, i.e. 1, 2, 3, 5 are also present here. The obvious difference between the two patterns is in the properties of high order parastichies and is particularly evident in the relationship between 3- and 8-parastichies: they are of the same chirality in the Fibonacci pattern, whereas they oppose each other in the pattern shown on the picture. This fact is probably responsible for not recognizing the 3:8 pattern in plants with a low expression of phyllotaxis in the shoot. On the basis of low order parastichies such as 1:2, 2:3 or 3:5 it is likely to be erroneously classified as the main Fibonacci pattern. Indeed, as Roberts (1984) points out, the data on the divergence angle in plants provided by Fujita (1939) indicate a slightly elevated frequency of patterns with the angle averaging 132 degrees. Another question, beside its recognition, is the stability of the 3:8 pattern. In most cases when it was encountered in *Magnolia*, it was involved in transformation (see Fig. 7). Closer analysis of the uppermost primordia indicates that even the very clear pattern shown on this picture is about to transform into another, yet undetectable (most probably 4:7 or 4:8). The uppermost primordia are numbered. The course of 11-parastichies is shown with a broken line.

Fig. 11. *Magnolia acuminata* – SEM image of another rare pattern regarded sometimes, similarly as that shown on Fig. 10, as the pattern „that cannot exist”. However, the analysis of the geometric relations shows that this pattern expressed as 7:10 should not be very different from the rather common Lucas pattern expressed as 7:11 (to which in turn the 8:11 expression should be very close). In fact there is a possibility, identically as in the case of the 1, 3, 8... and the main Fibonacci patterns, that in plants with a low expression of phyllotaxis, the 1, 3, 7... pattern is mistakenly qualified as the Lucas. It is because the 4-parastichies connecting every 4-th primordium also exist here. What makes a difference is their orientation. Here they are inclined in the same direction as 7-parastichies, whereas in the Lucas pattern they oppose each other. The uppermost primordia are numbered. The course of 10-parastichies is shown with a broken line.

Fig. 12. *Magnolia acuminata* – SEM image of the apex with a very regular trijugate pattern. The process of organogenesis is well advanced – the diameter of the unused portion of the apex surface is small. The carpel primordia are initiated in 3-merous whorls. Each whorl is rotated in respect to the previous one by an angle $137,5/3$. This is why the ortostichies do not form as in typically whorled patterns. The parastichy numbers belong to the phyllotactic series, in which the consecutive members are the numbers of the main Fibonacci series multiplied by 3. There are three ontogenetic helices – each primordium of one whorl is positioned on one of them. The pattern, extremely rare in conifers, is quite frequent in magnolia gynoecia (see also Fig. 16). On the level of 3:6 expression it makes, together with the previous two patterns, an elegant, smooth sequence of changing contacts between primordia: $3:6 \rightarrow 3:7 \rightarrow 3:8$. This is yet another reason for accepting the existence of the "impossible" patterns. The primordia of the three uppermost whorls are marked. These, that belong to one whorl bear the same number. The course of 9-parastichies is shown with a broken line.

Fig. 13. *Magnolia acuminata* – floral apex with a clear dislocation in the pattern of S-parastichies. This is the same apex as that shown on Fig. 8C but seen from above. Atop of the dislocation carpel primordia fuse. Phyllotaxis changes from simple (7:9) to multiple (6:9). In the upper portion of the system, above the dislocation, the primordia appear in a groups of three (marked with the same symbol), which is typical of trigyny. The chirality of primordia initiation i.e. the direction of the ontogenetic helix alters as a result of this transition. The center of the apex, calculated for each pattern separately, shifts when phyllotaxis changes (black dots).

Fig. 14. *Magnolia acuminata* – floral apex with a sectorial (localized) disturbance in the distribution of the uppermost primordia. It is interpreted as transformation of the second accessory (5:9) into the main Fibonacci pattern (5:8) currently taking place. The course of 9-parastichies, shown with a broken line becomes irregular in one sector of circumference because the youngest primordia are still numbered according to the 5:9 pattern. One of these parastichies ends its course.

structures becomes highly improbable. The quantitative analysis performed on the elongated gynoecia has the advantage of being fast. The scrutiny of a large quantity of shoots in a short time is possible. This method demonstrated that there are differences in the frequency of various patterns between species and even individual trees. The dogma of the Fibonacci pattern prevalence does not hold any longer since trees have been found in which the bijugy or trijugy dominates (Fig. 16). A variety of phyllotactic patterns is present in the gynoecia of all examined species of *Magnolia*. However, in *M. acuminata* the diversity is much greater than in any other species. Thus the most suitable object for studies on the phenomenon of phyllotaxis diversity has been found! It is highly recommended for further studies on what is possible and what is forbidden in phyllotaxis. They are particularly important since there is a theoretical model, that excludes two of the patterns shown here (Jean 1992, 1994). In the floral meristems of *Magnolia*, various phyllotactic patterns and transformations are extremely frequent, much more than in coniferous vegetative shoots. It is interesting, that tetrajugy, which is quite frequent in conifers, is very rare in *Magnolia*, whereas trijugy – extremely rare in conifers, is in *Magnolia* common. The frequency of various patterns is not the same for different *Magnolia* species. There are even differences between individual trees. This suggests that genetic factors might be involved.

The reason for such unusual behavior of *Magnolia* is unknown. The proposed explanation refers to the idea put forward by geneticists studying volatile sectorial chimeras in *Juniperus* (Ruth et al. 1985, Klekowski 1988). It is the idea of stochastic meristems with impermanent initials, where a specific mechanism selects out mutations, either on the level of a cell population composing one particular meristem or on the level of a population of meristems in the plant. The significance of this discovery cannot be underestimated: this strategy protects the initial (zygotic) identity of the genetic information carried by the initial cells which divide constantly for a long time. Multiple replications of DNA, especially in long living plants such as trees, represent a serious threat of somatic mutations accumulating. There is however yet another, very interesting implication of this phenomenon. The diplontic selection occurring in the stochastic meristems is a critical moment in the apex ontogeny. Selecting initials anew from the population of cells occupying the distal portion of the apex, results undoubtedly in a displacement of the growth center. With the change of the vertex position, the axis of the meristem changes its orientation becoming slightly inclined. The cells located along the circumference of the apex are now differently positioned with respect to the existing until now trajectories of the principal directions of growth (Hejnowicz 1984). Recognizing it, they either speed up or slow down their division activity. This in turn creates local instability of apex circumference – uneven expansion and/or a local deficit of the surface. The immediate morphogenetic effect is a dislocation and the alteration of phyllotaxis. The simulation of this process based on growth tensor analysis, but including also patterns of cell divisions has already been performed successfully (Nakielski, Zagórska-Marek, unpublished).

Is there any evidence of axial shift and circumferential change in real apices? First of all, it can be demonstrated with a computer program, which calculates the center of the apical dome from the positions of primordia, so that there is a measurable shift of the center when phyllotaxis changes (Fig. 13). Secondly, dislocation itself represents a strictly localized disturbance of the pattern. In addition the primordia fusion situated (if present) always atop of the dislocation, where one

of the parastichies ends its course, seems to indicate a local surface deficit in the organogenic region of the apex. Being already initiated, primordia fuse, as if there was not enough space for them to develop as discrete structures. An even more important implication is that primordia can be initiated circumferentially below(!) the critical distance between them. This might be an evidence of a vertical factor being more important in controlling the initiation process. It could be a vascular system as postulated by Larson (1980) in result of his studies on the phyllotaxis and the vascular system in *Populus deltoides*.

Plants probably differ in geometric stability of their meristems. It is easy to accept the hypothesis, that the more pronounced the stochastic character of the meristem is, the more likely becomes its ultimate effect, i.e. phyllotactic transformation. However, the change in apex circumference does not always have to be sufficient to accommodate new parastichy or to cause its loss. This might be especially true for meristems with big primordia, i. e. with a high ratio of primordia size to the apex size. Nevertheless, when the primordia are relatively small as in coniferous apices or in developing *Magnolia* flowers, the phyllotactic pattern should be affected.

The size of primordia relative to the apex size is most likely determined genetically. This would explain why some plants as *Torreya* or *Cephalotaxus* have exclusively bijugate phyllotaxis. According to the author's unpublished results, in both these genera leaf primordia are much larger than in *Picea* or in *Abies*. Most of the plants have large primordia and small meristems. Looking at the left corner of the phyllotactic grid (Fig. 2) we can easily understand why in such a case there are only a few phyllotactic options: the main Fibonacci pattern expressed as 1:2 or 2:3, the decussate pattern as 2:2, the bijugy as 2:4, the tricusate pattern as 3:3 and the Lucas pattern as 3:4. These are indeed the most common patterns of phyllotaxis occurring in plants. It should be emphasized that the main Fibonacci has twice as many chances to develop under such circumstances as any other pattern. Its prevalence becomes understandable. Moreover, the grid demonstrates that under the rule of minimal change, the only way to spirality from the opposite arrangement of cotyledones leads, in dicots ontogeny, through the Fibonacci pattern: 2:2 → 2:3 or 2:2 → 1:2.

As long as the further changes of apex circumference are distributed evenly during the continuous transition (Zagórska-Marek 1987), there is no change in the quality of phyllotaxis, only the expression of the pattern becomes higher or lower. In the heads of *Asteraceae* exemplified by *Helianthus* such a process increases the expression of the main Fibonacci pattern to 34:55 or higher. This way the continuous transitions contribute to the maintenance and thus to the high frequency of the main Fibonacci pattern in plants.

In conclusion: the stochastic character of the meristem and rather small primordia on a large apex constitute a prescription for rich phyllotactic diversity of patterns. It is assumed, that *Magnolia acuminata* is perfect in meeting these two requirements.

Many ideas presented above are hypothetical and should be treated as possible inspiration for further inquiries. If dislocations are more likely to occur in geometrically unstable meristems with impermanent initials, the clonal structure (cell packets) of the apical dome surface should reflect this instability. This analysis is currently under way.

Fig. 15. The frequency of the phyllotactic patterns in different *Magnolia* species. In all the trees of these species the main Fibonacci pattern is dominant. In *M. salicifolia* (the same tree is shown in two consecutive years) the other most frequent pattern is the Lucas pattern (4:7) whereas in other species it is the main bijugy (4:6). In *M. stellata* the frequency of the main Fibonacci pattern is lowered and of the less common patterns – slightly elevated. In *M. denudata* the variety of patterns is even greater though the Fibonacci pattern is still dominant. Below the species name (upper right corner of each diagram) is the tree location (PL – Poland, USA – The United States) and the year of collecting the shoots from the tree. In the last row there is a total number of shoots. The sequence of patterns as well as their expressions are established arbitrarily. Black and white bars represent respectively the S and Z chiral configuration of each pattern.

Fig. 16. The frequency of the phyllotactic patterns in different trees of *Magnolia acuminata*. The frequencies of all patterns are relatively high at the expense of these generally regarded as the most common. The main Fibonacci pattern dominates significantly only in one tree (middle right). In other tree the trijugate pattern (6:9) is most frequent (bottom right), in yet another (bottom left) the main bijugy (4:6). In these two trees the main Fibonacci is not even a second most frequent pattern. Further description as in Fig. 15.

Fig. 17. The frequency of various types of phyllotactic transitions in two trees of *M. acuminata*. They differ one from another in a type of most frequent transition. On the left side of each diagram the initial patterns are listed vertically. At the level of each of them the horizontal bar is drawn, in which the ultimate pattern is given. The length of the bar reflects the frequency of the particular transition. The orientation of parastichies in the initial and ultimate pattern is sequentially the same, i.e. the first number of each pair refers to parastichies oriented in the same direction.

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LITERATURE CITED

- ADLER I., 1974. A model of contact pressure in phyllotaxis. *J. Theor. Biol.* 45: 1-79.
- BATTEY N.H., NICHOLLS D.R.A., POUTEAU S.A., COEN E.S., 1993. Apical structure and development on flowering and reversion. In: Abstracts of The XV International Botanical Congress, Yokohama, Japan.
- BRAVAIS L., BRAVAIS A., 1837. Essai sur la disposition des feuilles curvisériées. *Ann. Sci. Nat. Bot. Vég.* 7-8.
- BRAVAIS L., BRAVAIS A., 1839. Essai sur la disposition générale des feuilles rectisériées. *Ann. Sci. Nat. Bot.* 12.
- CAMEFORT H., 1956. Étude de la structure du point végétatif et des variations phyllotaxiques chez quelques gymnospermes. *Ann. Sci. Bot.* 17: 1-185.
- CUTTER E.G., VOELLER B.R., 1959. Changes in leaf arrangement in individual fern apices. *J. Linn. Bot.* 56: 225-236.
- CANNELL M.G.R., BOWLER K.C., 1978. Phyllotactic arrangements of needles on elongating conifer shoots: a computer simulation. *Can. J. Forest Res.* 8: 138-141.
- CHARLTON W. A., 1994. The rotated-lamina syndrom. IV. Relationships between rotation and symmetry in *Magnolia* and other cases. *Can. J. Bot.* 72: 25-38.
- DAVIS T.A., BOSE T.K., 1971. Fibonacci systems in aroids. *Fib.Q.* 9: 253-263.
- ERBAR C., LEINS P., 1982. Zur Spirale in Magnolien-Blüten. *Beitr. Biol. Pflanzen* 56: 225-241.
- ERBAR C., LEINS P., 1983. Zur Sequenz von Blütenorganen bei einigen Magnoliiden. *Bot. Jahrb. Syst.* 103: 433-449.
- FUJITA T., 1937. Über die Reihe 2, 5, 7, 12... in der schraubigen Blattstellung und die mathematische Betrachtung verschiedener Zahlenreihensysteme. *Bot. Mag. Tokyo* 51: 480-489.
- FUJITA T., 1939. Statistische Untersuchungen über den Divergenzwinkel bei den schraubigen Organstellungen. *Bot. Mag. Tokyo* 53: 194-199.
- GÓMEZ-CAMPO C., 1974. Phyllotactic patterns in *Bryophyllum tubiflorum*. *Harv. Bot. Gaz.* 135: 49-58.
- GREGORY R.A., ROMBERGER J.A., 1972. The shoot apical ontogeny of the *Picea abies* seedling. I. Anatomy, apical dome diameter, and plastochron duration. *Am. J. Bot.* 59: 587-597.
- HEJNOWICZ Z., 1984. Trajectories of principal directions of growth, natural coordinate system in growing plant organ. *Acta Soc. Bot. Poloniae* 53: 29-42.
- JEAN R.V., 1992. Model testing in phyllotaxis. *J. Theor. Biol.* 156: 41-52.
- JEAN R.V., 1994. Phyllotaxis. A Systemic Study in Plant Morphogenesis. Cambridge Univ. Press, Cambridge, New York, Melbourne.
- JENSEN L.C., 1968. Primary stem vascular patterns in three subfamilies of the *Crassulaceae*. *Am. J. Bot.* 55: 553-563.
- KLEKOWSKI E.J. Jr., 1988. Mutation, developmental selection, and plant evolution. Columbia Univ. Press, New York.
- KWIATKOWSKA D., 1993. Ontogenetic changes of phyllotaxis and primary vasculature in *Anagallis arvensis*. In: Abstracts of The XV International Botanical Congress, Yokohama, Japan.
- KWIATKOWSKA D., 1994. Zmiany rozwojowe wzoru filotaksji i pierwotnego systemu waskularnego pędów *Anagallis arvensis*. PhD Thesis. Wrocław University.
- LARSON P.R., 1980. Interrelations between phyllotaxis, leaf development and the primary-secondary vascular transition in *Populus deltoides*. *Ann. Bot.* 46: 757-769.
- NOOTEBOOM H.P., 1985. Notes on *Magnoliaceae*. *Blumea* 31: 65-121.
- NOOTEBOOM H.P., 1987. Notes on *Magnoliaceae*. II. *Blumea* 32: 343-382.

- NOOTEBOOM H.P., 1993. *Magnoliaceae*. In: The Families and Genera of Vascular Plants. Vol.II: Flowering Plants – Dicotyledons. K. Kubitzki (Ed.), pp. 391-401. Springer-Verlag, Berlin, Heidelberg.
- ROBERTS D.W., 1984. A chemical contact pressure of phyllotaxis. *J. Theor. Biol.* 108: 481-490.
- ROMBERGER J.A., HEJNOWICZ Z., HILL J.F., 1993. *Plant Structure: Function and Development*. Springer Verlag, Berlin, New York.
- RUTH J., KLEKOWSKI E.J., STEIN O.L., 1985. Impermanent initials of the shoot apex and diplotic selection in a *Juniperus* chimera. *Amer. J. Bot.* 71: 1127-1135.
- TUCKER S.C., 1960. Ontogeny of the floral apex of *Michelia fuscata*. *Amer. J. Bot.* 47: 266-277.
- TUCKER S.C., 1961. Phyllotaxis and vascular organization of the carpels in *Michelia fuscata*. *Am. J. Bot.* 48: 60-71.
- ZAGÓRSKA-MAREK B., 1985. Phyllotactic patterns and transitions in *Abies balsamea*. *Can. J. Bot.* 63: 1844-1854.
- ZAGÓRSKA-MAREK B., 1987. Phyllotaxis triangular unit; phyllotactic transitions as the consequences of the apical wedge disclinations in a crystal-like pattern of the units. *Acta Soc. Bot. Poloniae* 56: 229-255.
- ZAGÓRSKA-MAREK B., 1993. Nowe spojrzenie na symetrię wzorów ulistnienia. In: *Symetrie w naukach przyrodniczych*. T.II pp. 239-256. Wrocław University (in Polish).

RÓŻNORODNOŚĆ FILOTAKSJII W KWIATACH MAGNOLII

STRESZCZENIE

Zbadano wzory filotaksji występujące w słupkowi sześciu gatunków magnolii: *M. acuminata*, *M. denudata*, *M. salicifolia*, *M. soulangeana*, *M. stellata* i *M. virginiana*. Stwierdzono wielką różnorodność wzorów. Znalezione między innymi niezmiernie rzadkie filotaksje wykluczane przez niektóre modele teoretyczne. Potwierdzono występowanie zjawiska ontogenetycznych zmian filotaksji, które dokonują się poprzez pojawianie się dyslokacji w periodycznym układzie elementów słupkowi. Wymienioną różnorodność, którą najłatwiej stwierdzić analizując pędy generatywne zaraz po kwitnieniu, potwierdzono analizą struktury merystemu kwiatowego przeprowadzoną w mikroskopie elektronowym skaningowym. W badaniach tych zastosowano metodę sporządzania odcisków reprosilowych i replik epoksydowych. W poszukiwaniu przyczyn różnorodności, której wyraźne stopniowanie zauważono w obrębie rodzaju, postawiono hipotezę, że jedną z nich może być stochastyczność merystemu oraz okresowe przemieszczanie się centrum wzrostowego wierzchołka.

SŁOWA KLUCZOWE: magnolia, filotaksja, merystem apikalny, dyslokacje, sieć krystaliczna, morfogeneza roślin.